LABORATORY OF BIOCHEMICAL GENETICS

Neuroblastoma and related lines of cultured cells were used as model systems for studies on synapse formation and other aspects of neuronal development. Prolonged elevation of cyclic AMP levels of neuroblastoma cells has profound, long-lived, stimulatory effects on the ability of the cells to form synapses and transmit information to other cells, which is due, in part, to the appearance of functional voltage-sensitive calcium channels. cDNA and genomic DNA clones were obtained that correspond to the α-subunit of L-type voltage-sensitive calcium channels of rat brain. In addition, seventeen cDNA clones were obtained that correspond to species of RNA that increase in abundance when neuroblastoma-glioma hybrid cells are treated with dibutyryl cAMP. Two of the cDNA clones were identified by nucleotide sequence analysis. Clone pNG-10 DNA corresponds to RNA transcribed from the light strand of mitochondrial DNA that functions as an RNA primer needed for the initiation of synthesis of heavy strand mitochondrial DNA. This species of RNA increases 40-fold in response to dibutyryl cAMP. Clone NG-32 DNA corresponds to mRNA for ATP synthase subunit 6, which is transcribed from a heavy strand mitochondrial gene and codes for a protein that is part of the H^+ channel of the ATP synthase complex. This species of mRNA increases 8-fold in response to dibutyryl cAMP. These results show that RNA transcripts from both light and heavy mitochondrial DNA strands increase in abundance when NG108-15 cells are treated with dibutyryl cAMP. The possibility that cAMP regulates the replication of mitochondrial DNA or the ability of mitochondria to synthesize ATP are problems for future studies.

Four novel Drosophila homeobox genes were cloned and partially sequenced. The homeobox family of genes code for proteins that regulate the expression of genes during development, and some ~~of the known~~ Drosophila homeobox genes are known to regulate pathways of differentiation. Three of the 4 new homeobox genes, NK-1, NK-3, and NK-4, were mapped to the 93 E region of the right arm of the third chromosome. The fourth novel homeobox gene was mapped to the 1C region of the sex chromosome. The NK-1 gene is expressed in embryos 3 to 12 hours after fertilization. Nucleotide sequence analysis showed that the NK-1 gene has 3 exons, and that 1 of the 2 introns detected resides within the homeobox region. These genes provide an experimental system that can be used to define the mechanisms that regulate the expression of these homeobox genes as well as the regulatory effects of the homeobox protein products ~~of the genes~~ on the expression of other genes.